Connecting via Winsock to STN

Welcome to STN International! Enter x:x

LOGINID:SSSPTA1644PNH

PASSWORD:
TERMINAL (ENTER 1, 2, 3, OR ?):2

* * * * * * * * * * Welcome to STN International * * * * * * * * * *

NEWS 1		Web Page URLs for STN Seminar Schedule - N. America
NEWS 2	Jan 25	BLAST(R) searching in REGISTRY available in STN on the Web
NEWS 3	Jan 29	FSTA has been reloaded and moves to weekly updates
NEWS 4	Feb 01	DKILIT now produced by FIZ Karlsruhe and has a new update frequency
NEWS 5	Feb 19	Access via Tymnet and SprintNet Eliminated Effective 3/31/02
NEWS 6	Mar 08	Gene Names now available in BIOSIS
NEWS 7	Mar 22	TOXLIT no longer available
NEWS 8	Mar 22	TRCTHERMO no longer available
NEWS 9	Mar 28	US Provisional Priorities searched with P in CA/CAplus and USPATFULL
NEWS 10	Mar 28	LIPINSKI/CALC added for property searching in REGISTRY
NEWS 11	Apr 02	PAPERCHEM no longer available on STN. Use PAPERCHEM2 instead.
NEWS 12	Apr 08	"Ask CAS" for self-help around the clock
NEWS 13	Apr 09	BEILSTEIN: Reload and Implementation of a New Subject Area
NEWS 14	Apr 09	ZDB will be removed from STN
NEWS 15	Apr 19	US Patent Applications available in IFICDB, IFIPAT, and IFIUDB
NEWS 16	Apr 22	Records from IP.com available in CAPLUS, HCAPLUS, and ZCAPLUS
NEWS 17	Apr 22	BIOSIS Gene Names now available in TOXCENTER
NEWS 18	Apr 22	Federal Research in Progress (FEDRIP) now available
NEWS 19	Jun 03	New e-mail delivery for search results now available
NEWS 20	Jun 10	MEDLINE Reload
NEWS 21	Jun 10	PCTFULL has been reloaded
NEWS 22	Jul 02	FOREGE no longer contains STANDARDS file segment

NEWS EXPRESS	February 1 CURRENT WINDOWS VERSION IS V6.0d, CURRENT MACINTOSH VERSION IS V6.0a(ENG) AND V6.0Ja(JP), AND CURRENT DISCOVER FILE IS DATED 05 FEBRUARY 2002
NEWS HOURS	STN Operating Hours Plus Help Desk Availability
NEWS INTER	General Internet Information
NEWS LOGIN	Welcome Banner and News Items
NEWS PHONE	Direct Dial and Telecommunication Network Access to STN
NEWS WWW	CAS World Wide Web Site (general information)

Enter NEWS followed by the item number or name to see news on that specific topic.

* * * * * * * * * * * * * * * STN Columbus * * * * * * * * * * * * * * *

```
FILE 'HOME' ENTERED AT 16:35:22 ON 18 JUL 2002

=> file medline embase biosis scisearch caplus
COST IN U.S. DOLLARS                                    SINCE FILE      TOTAL
                                                           ENTRY    SESSION
FULL ESTIMATED COST                                         0.21       0.21

FILE 'MEDLINE' ENTERED AT 16:35:40 ON 18 JUL 2002

FILE 'EMBASE' ENTERED AT 16:35:40 ON 18 JUL 2002
COPYRIGHT (C) 2002 Elsevier Science B.V.  All rights reserved.

FILE 'BIOSIS' ENTERED AT 16:35:40 ON 18 JUL 2002
COPYRIGHT (C) 2002 BIOLOGICAL ABSTRACTS INC.(R)

FILE 'SCISEARCH' ENTERED AT 16:35:40 ON 18 JUL 2002
COPYRIGHT (C) 2002 Institute for Scientific Information (ISI) (R)

FILE 'CAPLUS' ENTERED AT 16:35:40 ON 18 JUL 2002
USE IS SUBJECT TO THE TERMS OF YOUR STN CUSTOMER AGREEMENT.
PLEASE SEE "HELP USAGETERMS" FOR DETAILS.
COPYRIGHT (C) 2002 AMERICAN CHEMICAL SOCIETY (ACS)

=> s protein aggregration
   4 FILES SEARCHED...
L1             1 PROTEIN AGGREGRATION

=> d l1 cbib abs

L1   ANSWER 1 OF 1  CAPLUS  COPYRIGHT 2002 ACS
1978:421199   Document No. 89:21199  The effect of in vivo aging of the human
     erythrocyte on the proteins of the plasma membrane.  A comparison with
     metabolic depletion and blood bank storage.  Kadlubowski, Michael (Dep.
     Physiol., Univ. St. Andrews, St. Andrews/Fife, Scot.).  Int. J. Biochem.,
     9(2), 79-88 (English) 1978.  CODEN: IJBOBV.  ISSN: 0020-711X.
AB   The effect on the proteins of the human erythrocyte plasma membrane of in
     vivo aging and in vitro metabolic depletion by methods including glucose
     depletion and use of F- and iodoacetate were compared and a marked
     similarity was found.  This enabled a mechanistic explanation for some of
     the changes to be advanced.  Protein 2 appears to be highly susceptible to
     the products of lipid peroxidn., aggregating to yield the very
     high-mol.-wt. material in the 0.5 region.  The increase in protein 2.3 may
     be the result of a Ca-induced aggregation of .gtoreq.1 of the lower
     mol.-wt. proteins.  The decrease in membrane integrity which leads to the
     loss of protein 7 is probably the result of mech. stresses, oxidative
     denaturation of the membrane components, and failure of the repair
     mechanisms.  The accretion of proteins 4.2, 5 and 6 and Hb from the
     cytoplasm to the membrane is undoubtedly a result of age-related metabolic
     incompetence.  However, the chain of events linking the latter to the
     eventual binding of the proteins to the membrane is unclear and likely to
     be highly complex.  The direct involvement of ATP and Ca is apparently not
     of any great significance.


=> s amyloid forming protein
   3 FILES SEARCHED...
L2            50 AMYLOID FORMING PROTEIN

=> s l2 and inhibitor
L3             5 L2 AND INHIBITOR

=> dup remove l3
PROCESSING COMPLETED FOR L3
```

L4 1 DUP REMOVE L3 (4 DUPLICATES REMOVED)

=> d l4 cbib abs

L4 ANSWER 1 OF 1 MEDLINE DUPLICATE 1
94151341 Document Number: 94151341. PubMed ID: 8108423. Increased body temperature accelerates aggregation of the Leu-68-->Gln mutant cystatin C, the **amyloid-forming protein** in hereditary cystatin C amyloid angiopathy. Abrahamson M; Grubb A. (Department of Clinical Chemistry, University of Lund, University Hospital, Sweden.) PROCEEDINGS OF THE NATIONAL ACADEMY OF SCIENCES OF THE UNITED STATES OF AMERICA, (1994 Feb 15) 91 (4) 1416-20. Journal code: 7505876. ISSN: 0027-8424. Pub. country: United States. Language: English.

AB Hereditary cystatin C amyloid angiopathy is a dominantly inherited disorder, characterized by dementia, paralysis, and death from cerebral hemorrhage in early adult life. A variant of the cysteine proteinase **inhibitor**, cystatin C, is deposited as amyloid in the tissues of the patients and their spinal-fluid level of cystatin C is abnormally low. The disease-associated Leu-68-->Gln mutant (L68Q) cystatin C has been produced in an Escherichia coli expression system and isolated by use of denaturing buffers, immunosorption, and gel filtration. Parallel physicochemical and functional investigations of L68Q-cystatin C and wild-type cystatin C revealed that both proteins effectively inhibit the cysteine proteinase cathepsin B (equilibrium constants for dissociation, 0.4 and 0.5 nM, respectively) but differ considerably in their tendency to dimerize and form aggregates. While wild-type cystatin C is monomeric and functionally active even after prolonged storage at elevated temperatures, L68Q-cystatin C starts to dimerize and lose biological activity immediately after it is transferred to a nondenaturing buffer. The dimerization of L68Q-cystatin C is highly temperature-dependent, with a rise in incubation temperature from 37 to 40 degrees C resulting in a 150% increase in dimerization rate. The aggregation at physiological concentrations is likewise increased at 40 compared to 37 degrees C, by approximately 60%. These properties of L68Q-cystatin C have bearing upon our understanding of the pathophysiological process of hereditary cystatin C amyloid angiopathy. They might also be of clinical relevance, since medical intervention to abort febrile periods of carriers of the disease trait may reduce the in vivo formation of L68Q-cystatin C aggregates.

=> d his

(FILE 'HOME' ENTERED AT 16:35:22 ON 18 JUL 2002)

FILE 'MEDLINE, EMBASE, BIOSIS, SCISEARCH, CAPLUS' ENTERED AT 16:35:40 ON 18 JUL 2002

L1 1 S PROTEIN AGGREGRATION
L2 50 S AMYLOID FORMING PROTEIN
L3 5 S L2 AND INHIBITOR
L4 1 DUP REMOVE L3 (4 DUPLICATES REMOVED)

=> s l2 and minimize aggregation
L5 0 L2 AND MINIMIZE AGGREGATION

=> s l2 and aggregation
L6 14 L2 AND AGGREGATION

=> s l6 and inhibitor
L7 5 L6 AND INHIBITOR

=> dup remove l7
PROCESSING COMPLETED FOR L7
L8 1 DUP REMOVE L7 (4 DUPLICATES REMOVED)

=> d l8 cbib abs

L8 ANSWER 1 OF 1 MEDLINE DUPLICATE 1
94151341 Document Number: 94151341. PubMed ID: 8108423. Increased body temperature accelerates **aggregation** of the Leu-68-->Gln mutant cystatin C, the **amyloid-forming protein** in hereditary cystatin C amyloid angiopathy. Abrahamson M; Grubb A. (Department of Clinical Chemistry, University of Lund, University Hospital, Sweden.) PROCEEDINGS OF THE NATIONAL ACADEMY OF SCIENCES OF THE UNITED STATES OF AMERICA, (1994 Feb 15) 91 (4) 1416-20. Journal code: 7505876. ISSN: 0027-8424. Pub. country: United States. Language: English.

AB Hereditary cystatin C amyloid angiopathy is a dominantly inherited disorder, characterized by dementia, paralysis, and death from cerebral hemorrhage in early adult life. A variant of the cysteine proteinase **inhibitor**, cystatin C, is deposited as amyloid in the tissues of the patients and their spinal-fluid level of cystatin C is abnormally low. The disease-associated Leu-68-->Gln mutant (L68Q) cystatin C has been produced in an Escherichia coli expression system and isolated by use of denaturing buffers, immunosorption, and gel filtration. Parallel physicochemical and functional investigations of L68Q-cystatin C and wild-type cystatin C revealed that both proteins effectively inhibit the cysteine proteinase cathepsin B (equilibrium constants for dissociation, 0.4 and 0.5 nM, respectively) but differ considerably in their tendency to dimerize and form aggregates. While wild-type cystatin C is monomeric and functionally active even after prolonged storage at elevated temperatures, L68Q-cystatin C starts to dimerize and lose biological activity immediately after it is transferred to a nondenaturing buffer. The dimerization of L68Q-cystatin C is highly temperature-dependent, with a rise in incubation temperature from 37 to 40 degrees C resulting in a 150% increase in dimerization rate. The **aggregation** at physiological concentrations is likewise increased at 40 compared to 37 degrees C, by approximately 60%. These properties of L68Q-cystatin C have bearing upon our understanding of the pathophysiological process of hereditary cystatin C amyloid angiopathy. They might also be of clinical relevance, since medical intervention to abort febrile periods of carriers of the disease trait may reduce the in vivo formation of L68Q-cystatin C aggregates.

=> d his

(FILE 'HOME' ENTERED AT 16:35:22 ON 18 JUL 2002)

FILE 'MEDLINE, EMBASE, BIOSIS, SCISEARCH, CAPLUS' ENTERED AT 16:35:40 ON 18 JUL 2002

L1 1 S PROTEIN AGGREGRATION
L2 50 S AMYLOID FORMING PROTEIN
L3 5 S L2 AND INHIBITOR
L4 1 DUP REMOVE L3 (4 DUPLICATES REMOVED)
L5 0 S L2 AND MINIMIZE AGGREGATION
L6 14 S L2 AND AGGREGATION
L7 5 S L6 AND INHIBITOR
L8 1 DUP REMOVE L7 (4 DUPLICATES REMOVED)

=> dup remove l6
PROCESSING COMPLETED FOR L6
L9 6 DUP REMOVE L6 (8 DUPLICATES REMOVED)

=> d l9 1-6 cbib abs

L9 ANSWER 1 OF 6 MEDLINE DUPLICATE 1
2001361651 Document Number: 21315137. PubMed ID: 11423427. The assembly of amyloidogenic yeast sup35 as assessed by scanning (atomic) force

microscopy: an analogy to linear colloidal **aggregation**?. Xu S; Bevis B; Arnsdorf M F. (Department of Medicine and Molecular Genetics, The University of Chicago, Chicago, Illinois 60637, USA.. shxu@medicine.bsd.uchicago.edu) . BIOPHYSICAL JOURNAL, (2001 Jul) 81 (1) 446-54. Journal code: 0370626. ISSN: 0006-3495. Pub. country: United States. Language: English.

AB Amyloidosis is a class of diseases caused by protein **aggregation** and deposition in various tissues and organs. In this paper, a yeast **amyloid-forming protein** Sup35 was used as a model for understanding amyloid fiber formation. The dynamics of amyloid formation by Sup35 were studied with scanning force microscopy. We found that: 1) the assembly of Sup35 fibers begins with individual NM peptides that aggregate to form large beads or nucleation units which, in turn, form dimers, trimers, tetramers and longer linear assemblies appearing as a string of beads; 2) the morphology of the linear assemblies differ; and 3) fiber assembly suggests an analogy to the **aggregation** of colloidal particles. A dipole assembly model is proposed based on this analogy that will allow further experimental testing.

L9 ANSWER 2 OF 6 BIOSIS COPYRIGHT 2002 BIOLOGICAL ABSTRACTS INC.

2001:12173 Document No.: PREV200100012173. A census of glutamine/asparagine-rich regions: Implications for their conserved function and the prediction of novel prions. Michelitsch, Melissa D.; Weissman, Jonathan S. (1). (1) Department of Cellular and Molecular Pharmacology, University of California, 513 Parnassus Avenue, San Francisco, CA, 94143-0450: jsw1@itsa.ucsf.edu USA. Proceedings of the National Academy of Sciences of the United States of America, (October 24, 2000) Vol. 97, No. 22, pp. 11910-11915. print. ISSN: 0027-8424. Language: English. Summary Language: English.

AB Glutamine/asparagine (Q/N)-rich domains have a high propensity to form self-propagating amyloid fibrils. This phenomenon underlies both prion-based inheritance in yeast and **aggregation** of a number of proteins involved in human neurodegenerative diseases. To examine the prevalence of this phenomenon, complete proteomic sequences of 31 organisms and several incomplete proteomic sequences were examined for Q/N-rich regions. We found that Q/N-rich regions are essentially absent from the thermophilic bacterial and archaeal proteomes. Moreover, the average Q/N content of the proteins in these organisms is markedly lower than in mesophilic bacteria and eukaryotes. Mesophilic bacterial proteomes contain a small number (0-4) of proteins with Q/N-rich regions. Remarkably, Q/N-rich domains are found in a much larger number of eukaryotic proteins (107-472 per proteome) with diverse biochemical functions. Analyses of these regions argue they have been evolutionarily selected perhaps as modular "polar zipper" protein-protein interaction domains. These data also provide a large pool of potential novel prion-forming proteins, two of which have recently been shown to behave as prions in yeast, thus suggesting that **aggregation** or prion-like regulation of protein function may be a normal regulatory process for many eukaryotic proteins with a wide variety of functions.

L9 ANSWER 3 OF 6 SCISEARCH COPYRIGHT 2002 ISI (R)

2000:638452 The Genuine Article (R) Number: 317UV. Protein membrane interactions affect protein structure and **aggregation** in **amyloid-forming proteins**.. Good T (Reprint); Rymer D L. TEXAS A&M UNIV, COLLEGE STN, TX 77843. ABSTRACTS OF PAPERS OF THE AMERICAN CHEMICAL SOCIETY (26 MAR 2000) Vol. 219, Part 1, pp. 136-BIOT . Publisher: AMER CHEMICAL SOC. 1155 16TH ST, NW, WASHINGTON, DC 20036. ISSN: 0065-7727. Pub. country: USA. Language: English.

L9 ANSWER 4 OF 6 CAPLUS COPYRIGHT 2002 ACS

2000:327360 Protein membrane interactions affect protein structure and **aggregation** in **amyloid-forming proteins**.. Good, Theresa; Rymer, Dawn L. (Chemical Engineering,

Texas A&M University, College Station, TX, 77843-3122, USA). Book of Abstracts, 219th ACS National Meeting, San Francisco, CA, March 26-30, 2000, BIOT-136. American Chemical Society: Washington, D. C. (English) 2000. CODEN: 69CLAC.

AB At least sixteen different proteins can abnormally fold resulting in the formation of .beta.-sheet/ amyloid structure and disease. The prion protein, of transmissible spongiform encephalopathies, is one .beta.-sheet/**amyloid forming protein**. The conformational transition of the prion protein from an .alpha.-helical form, PrPC , to a predominantly .beta.-sheet form, PrPSC, is believed to be central to disease. We have evidence that interactions with phospholipid membranes may play a role in the PrPC to PrPSC conversion. Using a synthetic prion peptide, PrP(127-147), consisting of amino acids 127 to 147 of the human sequence, we have shown that peptide structure was stable under 25 solvation conditions with little sheet content. Upon incubation with phospholipid vesicles the peptide took on a substantial .beta.-sheet structure (>40%) which increased with time. We are examg. the role of membranes in conformational transitions of other **amyloid-forming proteins**. Our results will lead to insights into the mechanisms of amyloidosis.

L9 ANSWER 5 OF 6 BIOSIS COPYRIGHT 2002 BIOLOGICAL ABSTRACTS INC.

2000:177820 Document No.: PREV200000177820. Protein membrane interactions affect protein structure and **aggregation** in **amyloid-forming proteins**. Good, Theresa (1); Rymer, Dawn L. (1). (1) Chemical Engineering, Texas A and M University, 337 Zachry, College Station, TX, 77843-3122 USA. Abstracts of Papers American Chemical Society, (2000) Vol. 219, No. 1-2, pp. BIOT 136. Meeting Info.: 219th Meeting of the American Chemical Society. San Francisco, California, USA March 26-30, 2000 American Chemical Society. ISSN: 0065-7727. Language: English. Summary Language: English.

L9 ANSWER 6 OF 6 MEDLINE DUPLICATE 2

94151341 Document Number: 94151341. PubMed ID: 8108423. Increased body temperature accelerates **aggregation** of the Leu-68-->Gln mutant cystatin C, the **amyloid-forming protein** in hereditary cystatin C amyloid angiopathy. Abrahamson M; Grubb A. (Department of Clinical Chemistry, University of Lund, University Hospital, Sweden.) PROCEEDINGS OF THE NATIONAL ACADEMY OF SCIENCES OF THE UNITED STATES OF AMERICA, (1994 Feb 15) 91 (4) 1416-20. Journal code: 7505876. ISSN: 0027-8424. Pub. country: United States. Language: English.

AB Hereditary cystatin C amyloid angiopathy is a dominantly inherited disorder, characterized by dementia, paralysis, and death from cerebral hemorrhage in early adult life. A variant of the cysteine proteinase inhibitor, cystatin C, is deposited as amyloid in the tissues of the patients and their spinal-fluid level of cystatin C is abnormally low. The disease-associated Leu-68-->Gln mutant (L68Q) cystatin C has been produced in an Escherichia coli expression system and isolated by use of denaturing buffers, immunosorption, and gel filtration. Parallel physicochemical and functional investigations of L68Q-cystatin C and wild-type cystatin C revealed that both proteins effectively inhibit the cysteine proteinase cathepsin B (equilibrium constants for dissociation, 0.4 and 0.5 nM, respectively) but differ considerably in their tendency to dimerize and form aggregates. While wild-type cystatin C is monomeric and functionally active even after prolonged storage at elevated temperatures, L68Q-cystatin C starts to dimerize and lose biological activity immediately after it is transferred to a nondenaturing buffer. The dimerization of L68Q-cystatin C is highly temperature-dependent, with a rise in incubation temperature from 37 to 40 degrees C resulting in a 150% increase in dimerization rate. The **aggregation** at physiological concentrations is likewise increased at 40 compared to 37 degrees C, by approximately 60%. These properties of L68Q-cystatin C have bearing upon our understanding of the pathophysiological process of hereditary cystatin

C amyloid angiopathy. They might also be of clinical relevance, since medical intervention to abort febrile periods of carriers of the disease trait may reduce the in vivo formation of L68Q-cystatin C aggregates.

=> s human kappa IV light chain variable region
   3 FILES SEARCHED...
L10   0 HUMAN KAPPA IV LIGHT CHAIN VARIABLE REGION

=> s human kappa light chain
   3 FILES SEARCHED...
L11   198 HUMAN KAPPA LIGHT CHAIN

=> s l11 and protein aggregation
L12   0 L11 AND PROTEIN AGGREGATION

=> s serine protease inhibitor
L13   10620 SERINE PROTEASE INHIBITOR

=> s l13 and protein aggregation
L14   2 L13 AND PROTEIN AGGREGATION

=> dup remove l14
PROCESSING COMPLETED FOR L14
L15   2 DUP REMOVE L14 (0 DUPLICATES REMOVED)

=> 114

L14 IS NOT A RECOGNIZED COMMAND
The previous command name entered was not recognized by the system.
For a list of commands available to you in the current file, enter
"HELP COMMANDS" at an arrow prompt (=>).

=> d l15 1-2 cbib abs

L15 ANSWER 1 OF 2 SCISEARCH COPYRIGHT 2002 ISI (R)
2002:535685 The Genuine Article (R) Number: 567YJ. Association between conformational mutations in neuroserpin and onset and severity of dementia . Davis R L; Shrimpton A E; Carrell R W (Reprint); Lomas D A; Gerhard L; Baumann B; Lawrence D A; Yepes M; Kim T S; Ghetti B; Piccardo P; Takao M; Lacbawan F; Muenke M; Sifers R N; Bradshaw C B; Kent P F; Collins G H; Larocca D; Holohan P D. Univ Cambridge, Dept Haematol, Cambridge Inst Med Res, Cambridge, England (Reprint); Univ Cambridge, Dept Med, Cambridge Inst Med Res, Cambridge CB2 2QQ, England; Upstate Med Univ, Dept Pathol, Syracuse, NY USA; Univ Witten Herdecke, Inst Clin Neurosurg, Witten, Germany; Univ Magdeburg, Dept Psychiat, D-39106 Magdeburg, Germany; Amer Red Cross, Holland Lab, Dept Vasc Biol, Rockville, MD USA; Georgetown Univ, Med Ctr, Dept Neurol, Washington, DC 20007 USA; Yonsei Univ, Coll Med, Dept Pathol, Seoul, South Korea; Indiana Univ, Sch Med, Div Neuropathol, Alzheimer Dis Ctr, Indianapolis, IN USA; Childrens Natl Med Ctr, Dept Med Genet, Washington, DC 20010 USA; NHGRI, Med Genet Branch, NIH, Bethesda, MD 20892 USA; Baylor Coll Med, Dept Pathol, Houston, TX 77030 USA; Upstate Med Univ, Dept Neurol, Syracuse, NY USA; Upstate Med Univ, Dept Pharmacol, Syracuse, NY USA. LANCET (29 JUN 2002) Vol. 359, No. 9325, pp. 2242-2247. Publisher: LANCET LTD. 84 THEOBALDS RD, LONDON WC1X 8RR, ENGLAND. ISSN: 0140-6736. Pub. country: England; USA; Germany; South Korea. Language: English.
ABSTRACT IS AVAILABLE IN THE ALL AND IALL FORMATS
AB Background The aggregation of specific proteins is a common feature of the familial dementias, but whether the formation of neuronal inclusion bodies is a causative or incidental factor in the disease is not known. To clarify this issue, we investigated five families with typical neuroserpin inclusion bodies but with various neurological manifestations.

Methods Five families with neurodegenerative disease and typical neuronal inclusions had biopsy or autopsy material available for further examination. Immunostaining confirmed that the inclusions were formed of neuroserpin aggregates, and the responsible mutations in neuroserpin were identified by sequencing of the neuroserpin gene (SERPINI1) in DNA from blood samples or from extraction of histology specimens. Molecular modelling techniques were used to predict the effect of the gene mutations on three-dimensional protein structure. Brain sections were stained and the topographic distribution of the neuroserpin inclusions plotted.

Findings Each of the families was heterozygous for an aminoacid substitution that affected the conformational stability of neuroserpin. The least disruptive of these mutations (S49P), as predicted by molecular modelling, resulted in dementia after age 45 years, and presence of neuroserpin inclusions in only a few neurons. By contrast, the most severely disruptive mutation (G392E) resulted, at age 13 years, in progressive myoclonus epilepsy, with many inclusions present in almost all neurons.

Interpretation The findings provide evidence that inclusion-body formation is in itself a sufficient cause of neurodegeneration, and that the onset and severity of the disease is associated with the rate and magnitude of neuronal **protein aggregation**.

L15 ANSWER 2 OF 2 MEDLINE
2002366643 Document Number: 22106096. PubMed ID: 12112652. Familial conformational diseases and dementias. Crowther Damian C. (University of Cambridge Neurology Unit, Cambridge Institute for Medical Research, Cambridge, UK.) HUMAN MUTATION, (2002 Jul) 20 (1) 1-14. Journal code: 9215429. ISSN: 1098-1004. Pub. country: United States. Language: English.

AB Familial conformational diseases occur when a mutation alters the conformation of a protein resulting in abnormal intermolecular interactions, **protein aggregation**, and consequent tissue damage. The molecular mechanisms of conformational disease are best understood for the **serine protease inhibitor** (serpin) superfamily of proteins. The serpinopathies include alpha(1)-antitrypsin (SERPINA1) deficiency and the newly characterized familial encephalopathy with neuroserpin inclusion bodies (FENIB) resulting from mutations in the neuroserpin (SERPINI1) gene. This review discusses how insights gained from the study of the serpins may be used to guide our research into other common diseases such as Alzheimer disease, Huntington disease, and Parkinson disease.

=> d his

(FILE 'HOME' ENTERED AT 16:35:22 ON 18 JUL 2002)

FILE 'MEDLINE, EMBASE, BIOSIS, SCISEARCH, CAPLUS' ENTERED AT 16:35:40 ON 18 JUL 2002

L1	1	S PROTEIN AGGREGRATION
L2	50	S AMYLOID FORMING PROTEIN
L3	5	S L2 AND INHIBITOR
L4	1	DUP REMOVE L3 (4 DUPLICATES REMOVED)
L5	0	S L2 AND MINIMIZE AGGREGATION
L6	14	S L2 AND AGGREGATION
L7	5	S L6 AND INHIBITOR
L8	1	DUP REMOVE L7 (4 DUPLICATES REMOVED)
L9	6	DUP REMOVE L6 (8 DUPLICATES REMOVED)
L10	0	S HUMAN KAPPA IV LIGHT CHAIN VARIABLE REGION
L11	198	S HUMAN KAPPA LIGHT CHAIN
L12	0	S L11 AND PROTEIN AGGREGATION
L13	10620	S SERINE PROTEASE INHIBITOR
L14	2	S L13 AND PROTEIN AGGREGATION

L15 2 DUP REMOVE L14 (0 DUPLICATES REMOVED)

=> s l11 and beta amyloid
L16 0 L11 AND BETA AMYLOID

=> s l11 and protein folding
L17 5 L11 AND PROTEIN FOLDING

=> dup remove l17
PROCESSING COMPLETED FOR L17
L18 2 DUP REMOVE L17 (3 DUPLICATES REMOVED)

=> d l18 1-2 cbib abs

L18 ANSWER 1 OF 2 MEDLINE DUPLICATE 1
1998088448 Document Number: 98088448. PubMed ID: 9427009. X-ray crystallography reveals stringent conservation of protein fold after removal of the only disulfide bridge from a stabilized immunoglobulin variable domain. Uson I; Bes M T; Sheldrick G M; Schneider T R; Hartsch T; Fritz H J. (Institut fur Anorganische Chemie der Universitat, Gottingen, Germany.. uson@shelx.uni-ac.gwdg.de) . FOLDING AND DESIGN, (1997) 2 (6) 357-61. Journal code: 9604387. ISSN: 1359-0278. Pub. country: ENGLAND: United Kingdom. Language: English.

AB BACKGROUND: Immunoglobulin domains owe a crucial fraction of their conformational stability to an invariant central disulfide bridge, the closure of which requires oxidation. Under the reducing conditions prevailing in cell cytoplasm, accumulation of soluble immunoglobulin is prohibited by its inability to acquire and maintain the native conformation. Previously, we have shown that disulfide-free immunoglobulins can be produced in Escherichia coli and purified from cytoplasmic extracts. RESULTS: Immunoglobulin REIv is the variable domain of a **human kappa light chain**. The disulfide-free variant REIv-C23V/Y32H was crystallized and its structure analyzed by X-ray crystallography (2.8 A resolution). The conformation of the variant is nearly identical to that of the wild-type protein and the conformationally stabilized variant REIv-T39K. This constitutes the first crystal structure of an immunoglobulin fragment without a disulfide bridge. The lack of the disulfide bridge produces no obvious local change in structure (compared with the wild type), whereas the Y32H mutation allows the formation of an additional hydrogen bond. There is a further change in the structure that is seen in the dimer in which Tyr49 has flipped out of the dimer interface in the mutant. CONCLUSIONS: Immunoglobulin derivatives without a central disulfide bridge but with stringently conserved wild-type conformation can be constructed in a practical two-step approach. First, the protein is endowed with additional folding stability by the introduction of one or more stabilizing amino acid exchanges; second, the disulfide bridge is destroyed by substitution of one of the two invariant cysteines. Such derivatives can be accumulated in soluble form in the cytoplasmic compartment of the E. coli cell. Higher protein yields and evolutionary refinement of catalytic antibodies by genetic complementation are among the possible advantages.

L18 ANSWER 2 OF 2 MEDLINE DUPLICATE 2
97235016 Document Number: 97235016. PubMed ID: 9080189. Contribution of the intramolecular disulfide bridge to the folding stability of REIv, the variable domain of a human immunoglobulin kappa light chain. Frisch C; Kolmar H; Schmidt A; Kleemann G; Reinhardt A; Pohl E; Uson I; Schneider T R; Fritz H J. (Institut fur Molekulare Genetik, Georg-August-Universitat Gottingen, FRG.) FOLDING AND DESIGN, (1996) 1 (6) 431-40. Journal code: 9604387. ISSN: 1359-0278. Pub. country: ENGLAND: United Kingdom. Language: English.

AB BACKGROUND: Immunoglobulin domains contain about 100 amino acid residues folded into two beta-sheets and stabilized in a sandwich by a conserved

central disulfide bridge. Whether antibodies actually require disulfide bonds for stability has long been a matter of debate. The contribution made by the central disulfide bridge to the overall folding stability of the immunoglobulin REIv, the variable domain of a **human kappa light chain**, was investigated by introducing stabilizing amino acid replacements followed by removal of the disulfide bridge via chemical reduction or genetic substitution of the cysteine residues. RESULTS: Nine REIv variants were constructed by methods of protein engineering that have folding stabilities elevated relative wild-type REIv by (up to) 16.0 kJ mol-1. Eight of these variants can be cooperatively refolded after unfolding and chemical reduction of the disulfide bridge-in contrast to wildtype REIv. The stabilizing effect of one of these residue replacements (T39K) was rationalized by determining the structure of the respective REIv variant at 1.7 A. The loss of folding stability caused by reduction of the intramolecular disulfide bond is on average 19 kJ mol-1. Removal of the disulfide bridge by genetic substitution of C23 for valine resulted in a stable immunoglobulin domain in the context of the stabilizing Y32H amino acid exchange; again, REIv-C23V/Y32H has 18 kJ mol-1 less folding stability than REIv-Y32H. The data are consistent with the notion that all variants studied have the same overall three-dimensional structure with the disulfide bridge opened or closed. CONCLUSIONS: A comparison of the magnitude of the stabilizing effect exerted by the disulfide bond and the length of the mainchain loop framed by it suggests lowering of the entropy of the unfolded state as the sole source of the effect. Disulfide bonds are not necessary for proper folding of immunoglobulin variable domains and can be removed, provided the loss of folding stability is at least partly compensated by stabilizing amino acid exchanges.

```
=> s human immunoglobulin kappa light chain
   3 FILES SEARCHED...
L19         109 HUMAN IMMUNOGLOBULIN KAPPA LIGHT CHAIN

=> s l19 and protein folding
L20           4 L19 AND PROTEIN FOLDING

=> dup remove l20
PROCESSING COMPLETED FOR L20
L21           2 DUP REMOVE L20 (2 DUPLICATES REMOVED)

=> d l21 1-2 cbib abs

L21  ANSWER 1 OF 2     MEDLINE                                DUPLICATE 1
97235016  Document Number: 97235016.     PubMed ID: 9080189.   Contribution of
      the intramolecular disulfide bridge to the folding stability of REIv, the
      variable domain of a human immunoglobulin
      kappa light chain. Frisch C; Kolmar H; Schmidt
      A; Kleemann G; Reinhardt A; Pohl E; Uson I; Schneider T R; Fritz H J.
      (Institut fur Molekulare Genetik, Georg-August-Universitat Gottingen, FRG.
      ) FOLDING AND DESIGN, (1996) 1 (6) 431-40.  Journal code: 9604387. ISSN:
      1359-0278. Pub. country: ENGLAND: United Kingdom. Language: English.
AB    BACKGROUND: Immunoglobulin domains contain about 100 amino acid residues
      folded into two beta-sheets and stabilized in a sandwich by a conserved
      central disulfide bridge. Whether antibodies actually require disulfide
      bonds for stability has long been a matter of debate. The contribution
      made by the central disulfide bridge to the overall folding stability of
      the immunoglobulin REIv, the variable domain of a human kappa light chain,
      was investigated by introducing stabilizing amino acid replacements
      followed by removal of the disulfide bridge via chemical reduction or
      genetic substitution of the cysteine residues. RESULTS: Nine REIv variants
      were constructed by methods of protein engineering that have folding
      stabilities elevated relative wild-type REIv by (up to) 16.0 kJ mol-1.
```

Eight of these variants can be cooperatively refolded after unfolding and chemical reduction of the disulfide bridge-in contrast to wildtype REIv. The stabilizing effect of one of these residue replacements (T39K) was rationalized by determining the structure of the respective REIv variant at 1.7 A. The loss of folding stability caused by reduction of the intramolecular disulfide bond is on average 19 kJ mol-1. Removal of the disulfide bridge by genetic substitution of C23 for valine resulted in a stable immunoglobulin domain in the context of the stabilizing Y32H amino acid exchange; again, REIv-C23V/Y32H has 18 kJ mol-1 less folding stability than REIv-Y32H. The data are consistent with the notion that all variants studied have the same overall three-dimensional structure with the disulfide bridge opened or closed. CONCLUSIONS: A comparison of the magnitude of the stabilizing effect exerted by the disulfide bond and the length of the mainchain loop framed by it suggests lowering of the entropy of the unfolded state as the sole source of the effect. Disulfide bonds are not necessary for proper folding of immunoglobulin variable domains and can be removed, provided the loss of folding stability is at least partly compensated by stabilizing amino acid exchanges.

L21 ANSWER 2 OF 2 EMBASE COPYRIGHT 2002 ELSEVIER SCI. B.V.DUPLICATE 2
97003792 EMBASE Document No.: 1997003792. Structural and functional characterization of three **human immunoglobulin** .
kappa. light chains with different
pathological implications. Bellotti V.; Stoppino M.; Mangione P.P.; Fornasieri A.; Min L.; Merlini G.; Ferri G.. V. Bellotti, Department of Biochemistry, University of Pavia, Via Taramelli 3b, 27100 Pavia, Italy.
Biochimica et Biophysica Acta - Molecular Basis of Disease 1317/3 (161-167) 1996.
ISSN: 0925-4439. CODEN: BBADEX.
Publisher Ident.: S 0925-4439(96)00049-X. Pub. Country: Netherlands.
Language: English. Summary Language: English.
AB The structural properties of three immunoglobulins light chains: .kappa. SCI, responsible for light chain deposition disease (Bellotti, V., Stoppini, M., Merlini, G., Zapponi, M.C., Meloni, M.L., Banfi, G. and Ferri, G. (1991) Biochim. Biophys. Acta 1097, 177-182), k INC responsible for light chain amyloidosis (Ferri, G., Stoppini, M., Iadarola, P., Bellotti, V. and Merlini, G. (1989) Biochim. Biophys. Acta 995, 103-108) and the non-pathogenic .kappa. MOS were analyzed by fluorescence spectroscopy and circular dichroism. Comparative evaluation of the data shows that SCI and MOS have similar stability under different conditions, while the amyloid k INC behaves as a very unstable protein. As calculated from the GdnHCl curves, the midpoint of unfolding transition was 1.35 M for SCI, 1.20 M for MOS and 0.1 M for INC. Analysis of CD spectra evidences that the three proteins conserve their conformation in the range of pH 4-8. Change in temperature at pH 4.0 produces the premature transition of INC (T(m) 40.degree.C) with respect to SCI and MOS (T(m) 50.degree.C). At this pH both the pathological SCI and INC light chains aggregate at a temperature of 20.degree.C lower than the normal counterpart. The specific kidney deposition of .kappa. SCI has been evidenced after injection of the 125I labelled light chain into mice. No deposition was detectable in the case of INC and MOS.

```
=> s "BiP"
L22       5193 "BIP"

=> s l22 and "SMA aggregation"
L23          1 L22 AND "SMA AGGREGATION"

=> d l23 cbib abs

L23 ANSWER 1 OF 1 SCISEARCH COPYRIGHT 2002 ISI (R)
2001:201905 The Genuine Article (R) Number: 404RQ. Hsp70 and
```

antifibrillogenic peptides promote degradation and inhibit intracellular aggregation of amyloidogenic light chains. Dul J L; Davis D P; Williamson E K; Stevens F J; Argon Y (Reprint). Univ Chicago, Dept Pathol, 5841 S Maryland Ave, MC 1089, Chicago, IL 60637 USA (Reprint); Univ Chicago, Dept Pathol, Chicago, IL 60637 USA; Univ Chicago, Comm Immunol, Chicago, IL 60637 USA; Univ Chicago, Dept Mol Genet & Cell Biol, Chicago, IL 60637 USA; Argonne Natl Lab, Biosci Div, Argonne, IL 60439 USA. JOURNAL OF CELL BIOLOGY (19 FEB 2001) Vol. 152, No. 4, pp. 705-715. Publisher: ROCKEFELLER UNIV PRESS. 1114 FIRST AVE, 4TH FL, NEW YORK, NY 10021 USA. ISSN: 0021-9525. Pub. country: USA. Language: English.

ABSTRACT IS AVAILABLE IN THE ALL AND IALL FORMATS

AB In light chain (LC) amyloidosis an immunoglobulin LC assembles into fibrils that are deposited in various tissues. Little is known about how these fibrils form in vivo. We previously showed that a known amyloidogenic LC, SMA, can give rise to amyloid fibrils in vitro when a segment of one of its beta sheets undergoes a conformational change, exposing an Hsp70 binding site. To examine **SMA aggregation** in vivo, we expressed it and its wild-type counterpart, LEN, in COS cells. While LEN is rapidly oxidized and subsequently secreted, newly synthesized SMA remains in the reduced state. Most SMA molecules are dislocated out of the ER into the cytosol, where they are ubiquitinylated and degraded by proteasomes. A parallel pathway for molecules that are not degraded is condensation into perinuclear aggresomes that are surrounded by vimentin-containing intermediate filaments and are dependent upon intact microtubules. Inhibition of proteasome activity shifts the balance toward aggresome formation. Intracellular aggregation is decreased and targeting to proteasomes improved by overexpression of the cytosolic chaperone Hsp70. Importantly transduction into the cell of an Hsp70 target peptide, derived from the LC sequence, also reduces aggresome formation and increases SMA degradation. These results demonstrate that an amyloidogenic LC can aggregate intracellularly despite the common presentation of extracellular aggregates, and that a similar molecular surface mediates both in vitro fibril formation and in vivo aggregation. Furthermore, rationally designed peptides can be used to suppress this aggregation and may provide a feasible therapeutic approach.

```
=> s "SMA"
L24      18313 "SMA"

=> s l24 and binding protein
L25        185 L24 AND BINDING PROTEIN

=> s l25 and fibrils essembly
L26          0 L25 AND FIBRILS ESSEMBLY

=> s l25 and transthyretin
L27          0 L25 AND TRANSTHYRETIN

=> dup remove l25
PROCESSING COMPLETED FOR L25
L28          92 DUP REMOVE L25 (93 DUPLICATES REMOVED)

=> s l28 and hair pin loop
L29          0 L28 AND HAIR PIN LOOP

=> s l28 and endoplasmic reticulum chaperone
L30          0 L28 AND ENDOPLASMIC RETICULUM CHAPERONE

=> d his

     (FILE 'HOME' ENTERED AT 16:35:22 ON 18 JUL 2002)
```

FILE 'MEDLINE, EMBASE, BIOSIS, SCISEARCH, CAPLUS' ENTERED AT 16:35:40 ON 18 JUL 2002

L1 1 S PROTEIN AGGREGRATION
L2 50 S AMYLOID FORMING PROTEIN
L3 5 S L2 AND INHIBITOR
L4 1 DUP REMOVE L3 (4 DUPLICATES REMOVED)
L5 0 S L2 AND MINIMIZE AGGREGATION
L6 14 S L2 AND AGGREGATION
L7 5 S L6 AND INHIBITOR
L8 1 DUP REMOVE L7 (4 DUPLICATES REMOVED)
L9 6 DUP REMOVE L6 (8 DUPLICATES REMOVED)
L10 0 S HUMAN KAPPA IV LIGHT CHAIN VARIABLE REGION
L11 198 S HUMAN KAPPA LIGHT CHAIN
L12 0 S L11 AND PROTEIN AGGREGATION
L13 10620 S SERINE PROTEASE INHIBITOR
L14 2 S L13 AND PROTEIN AGGREGATION
L15 2 DUP REMOVE L14 (0 DUPLICATES REMOVED)
L16 0 S L11 AND BETA AMYLOID
L17 5 S L11 AND PROTEIN FOLDING
L18 2 DUP REMOVE L17 (3 DUPLICATES REMOVED)
L19 109 S HUMAN IMMUNOGLOBULIN KAPPA LIGHT CHAIN
L20 4 S L19 AND PROTEIN FOLDING
L21 2 DUP REMOVE L20 (2 DUPLICATES REMOVED)
L22 5193 S "BIP"
L23 1 S L22 AND "SMA AGGREGATION"
L24 18313 S "SMA"
L25 185 S L24 AND BINDING PROTEIN
L26 0 S L25 AND FIBRILS ESSEMBLY
L27 0 S L25 AND TRANSTHYRETIN
L28 92 DUP REMOVE L25 (93 DUPLICATES REMOVED)
L29 0 S L28 AND HAIR PIN LOOP
L30 0 S L28 AND ENDOPLASMIC RETICULUM CHAPERONE

=> s l25 and endoplasmic reticulum
L31 2 L25 AND ENDOPLASMIC RETICULUM

=> dup remove l31
PROCESSING COMPLETED FOR L31
L32 2 DUP REMOVE L31 (0 DUPLICATES REMOVED)

=> d 132 1-2 cbib abs

L32 ANSWER 1 OF 2 SCISEARCH COPYRIGHT 2002 ISI (R)
2001:201905 The Genuine Article (R) Number: 404RQ. Hsp70 and antifibrillogenic peptides promote degradation and inhibit intracellular aggregation of amyloidogenic light chains. Dul J L; Davis D P; Williamson E K; Stevens F J; Argon Y (Reprint). Univ Chicago, Dept Pathol, 5841 S Maryland Ave, MC 1089, Chicago, IL 60637 USA (Reprint); Univ Chicago, Dept Pathol, Chicago, IL 60637 USA; Univ Chicago, Comm Immunol, Chicago, IL 60637 USA; Univ Chicago, Dept Mol Genet & Cell Biol, Chicago, IL 60637 USA; Argonne Natl Lab, Biosci Div, Argonne, IL 60439 USA. JOURNAL OF CELL BIOLOGY (19 FEB 2001) Vol. 152, No. 4, pp. 705-715. Publisher: ROCKEFELLER UNIV PRESS. 1114 FIRST AVE, 4TH FL, NEW YORK, NY 10021 USA. ISSN: 0021-9525. Pub. country: USA. Language: English.
ABSTRACT IS AVAILABLE IN THE ALL AND IALL FORMATS
AB In light chain (LC) amyloidosis an immunoglobulin LC assembles into fibrils that are deposited in various tissues. Little is known about how these fibrils form in vivo. We previously showed that a known amyloidogenic LC, **SMA**, can give rise to amyloid fibrils in vitro when a segment of one of its beta sheets undergoes a conformational change, exposing an Hsp70 binding site. To examine **SMA** aggregation in vivo, we expressed it and its wild-type counterpart, LEN,

in COS cells. While LEN is rapidly oxidized and subsequently secreted, newly synthesized **SMA** remains in the reduced state. Most **SMA** molecules are dislocated out of the ER into the cytosol, where they are ubiquitinylated and degraded by proteasomes. A parallel pathway for molecules that are not degraded is condensation into perinuclear aggresomes that are surrounded by vimentin-containing intermediate filaments and are dependent upon intact microtubules. Inhibition of proteasome activity shifts the balance toward aggresome formation. Intracellular aggregation is decreased and targeting to proteasomes improved by overexpression of the cytosolic chaperone Hsp70. Importantly transduction into the cell of an Hsp70 target peptide, derived from the LC sequence, also reduces aggresome formation and increases **SMA** degradation. These results demonstrate that an amyloidogenic LC can aggregate intracellularly despite the common presentation of extracellular aggregates, and that a similar molecular surface mediates both in vitro fibril formation and in vivo aggregation. Furthermore, rationally designed peptides can be used to suppress this aggregation and may provide a feasible therapeutic approach.

L32 ANSWER 2 OF 2 SCISEARCH COPYRIGHT 2002 ISI (R)
92:605012 The Genuine Article (R) Number: JT720. EXPRESSION AND METHYLATION OF THE BETA-SUBUNIT GENE OF PROLYL 4-HYDROXYLASE - IN ERYTHROCYTES, TENDON AND CORNEA OF CHICK-EMBRYOS. CHIDA Y (Reprint); ISHIZAKI M; NAKAZAWA M; KAO W W Y. UNIV CINCINNATI, COLL MED, DEPT OPHTHALMOL, CINCINNATI, OH, 45267 (Reprint). CONNECTIVE TISSUE RESEARCH (1992) Vol. 28, No. 3, pp. 191-204. ISSN: 0300-8207. Pub. country: USA. Language: ENGLISH.
ABSTRACT IS AVAILABLE IN THE ALL AND IALL FORMATS

AB It has recently been demonstrated that the beta-subunit of prolyl 4-hydroxylase (E.C. 1.14.11.2) is die same gene product as protein disulfide isomerase (PDI) and cellular thyroid hormone **binding protein** (THP). Therefore, it is very likely that the beta-subunit of the prolyl 4-hydroxylase gene serves as a house keeping gene in most cell types. In the present study, we examined the distribution of the chicken beta-subunit of prolyl 4-hydroxylase/protein disulfide isomerase (CPHbeta/PDI) in erythrocytes, corneas and tendons of 13-, 17-, and 19-day-old chick embryos by immunohistochemistry using antibodies against CPHbeta/PDI. Our data indicate that erythrocytes do not express the CPHbeta/PDI gene whereas tendon cells express CPHbeta/PDI at all developmental stages examined. The basal cells of corneal epithelium express CPHbeta/PDI, but the superficial cell layers of stratified corneas of 19-day-old chick embryos do not. The expression of the CPHbeta/PDI gene is also confirmed by in situ hybridization with cDNA encoding CPHbeta/PDI. The results indicate that the expression of CPHbeta/PDI in cornea is probably developmentally regulated. It has been suggested that methylation of genomic DNA is one of many possible regulatory mechanisms for gene expression. In order to examine whether methylation of genomic DNA may play any role in die expression of the beta-subunit gene, genomic DNA was isolated from corneas, tendons, and erythrocytes of individual 13-, 17-, and 19-day-old chick embryos. DNA samples were digested with **Sma** I and Eco RI, or Pst I and **Sma** I and followed by either Msp I, Hpa II, or Hha I and were then subjected to Southern hybridization with P-32-labeled genomic DNA fragments of CPHbeta/PDI. Our results indicate that the CPHbeta/PDI gene is methylated at the Hha I site in the 4th exon in erythrocytes whereas the same sites in tendon and cornea are hypomethylated. Examination of 5'-end flanking sequences of exon 1 of the CPHbeta/PDI gene with the methylation sensitive endonucleases, Hha I and Hpa II did not reveal any difference in erythrocyte, cornea and tendon cells. Thus, our results indicated that DNA methylation may not play an important role in the expression or CPHbeta/PDI.

=> s transthyretin
L33 6819 TRANSTHYRETIN

=> s l33 and greek key
L34 1 L33 AND GREEK KEY

=> d l34 cbib abs

L34 ANSWER 1 OF 1 SCISEARCH COPYRIGHT 2002 ISI (R)
97:384557 The Genuine Article (R) Number: WY677. Function of the **Greek key** connection analysed using circular permutants of superoxide dismutase. Boissinot M; Karnas S; Lepock J R; Cabelli D E; Tainer J A; Getzoff E D; Hallewell R A (Reprint). UNIV LONDON IMPERIAL COLL SCI TECHNOL & MED, DEPT BIOCHEM, LONDON SW7 2AZ, ENGLAND (Reprint); UNIV LONDON IMPERIAL COLL SCI TECHNOL & MED, DEPT BIOCHEM, LONDON SW7 2AZ, ENGLAND; SCRIPPS CLIN & RES INST, DEPT BIOL MOL, LA JOLLA, CA 92037; BROOKHAVEN NATL LAB, DEPT CHEM, UPTON, NY 11973. EMBO JOURNAL (1 MAY 1997) Vol. 16, No. 9, pp. 2171-2178. Publisher: OXFORD UNIV PRESS. WALTON ST JOURNALS DEPT, OXFORD, ENGLAND OX2 6DP. ISSN: 0261-4189. Pub. country: ENGLAND; USA. Language: English.
ABSTRACT IS AVAILABLE IN THE ALL AND IALL FORMATS
AB Human Cu,Zn superoxide dismutase (SOD) is a single domain all beta-sheet protein with its eight beta-strands arranged as a **Greek key** beta-barrel or immunoglobulin fold. Three circularly permuted variants of SOD were made by joining the native amino- and carboxy-termini, and introducing new termini at sites originally within connections between beta-strands. The locations of the new termini were chosen to interrupt beta-turns between the two N-terminal beta-hairpins and the short cross-barrel **Greek key** connection, Expression levels in the Escherichia coli periplasm were indistinguishable from that of native SOD. Reaction rates for the purified proteins were similar to those of the native enzyme, indicating that the permutants are correctly folded, Interrupting the covalent cross-bracing provided by the **Greek key** connection reduced the stability of the protein by similar to 1.0 kcal/mol, indicating only a slight contribution to conformational stability. The experiments test and eliminate two hypotheses for folding pathways for **Greek key** beta-barrels that require N-terminal beta-hairpins or covalent attachment across the short **Greek key** connection.

=> s beta 2 microglobulin
L35 33245 BETA 2 MICROGLOBULIN

=> s l35 and protein assembly
L36 82 L35 AND PROTEIN ASSEMBLY

=> s l36 and inhibitor
L37 3 L36 AND INHIBITOR

=> dup remove l37
PROCESSING COMPLETED FOR L37
L38 3 DUP REMOVE L37 (0 DUPLICATES REMOVED)

=> d l38 1-3 cbib abs

L38 ANSWER 1 OF 3 EMBASE COPYRIGHT 2002 ELSEVIER SCI. B.V.
1998284351 EMBASE Assembly of MHC class I molecules with biosynthesized endoplasmic reticulum-targeted peptides is inefficient in insect cells and can be enhanced by protease **inhibitors**. Deng Y.; Gibbs J.; Bacik I.; Porgador A.; Copeman J.; Lehner P.; Ortmann B.; Cresswell P.; Bennink J.R.; Yewdell J.W.. Dr. J.W. Yewdell, Building 4, National Institute of Health, Bethesda, MD, United States. jyewdell@nih.gov. Journal of Immunology 161/4 (1677-1685) 15 Aug 1998.
Refs: 57.

ISSN: 0022-1767. CODEN: JOIMA3. Pub. Country: United States. Language: English. Summary Language: English.

AB To study the requirements for assembly of MHC class I molecules with antigenic peptides in the endoplasmic reticulum (ER), we studied Ag processing in insect cells. Insects lack a class I recognition system, and their cells therefore provide a 'blank slate' for identifying the proteins that have evolved to facilitate assembly of class I molecules in vertebrate cells. H-2Kb heavy chain, mouse **.beta.2-microglobulin**, and an ER-targeted version of a peptide corresponding to Ova257-264 were expressed in insect cells using recombinant vaccinia viruses. Cell surface expression of Kb-OVA257-264 complexes was quantitated using a recently described complex-specific mAb (25-D1.16). Relative to TAP-deficient human cells, insect cells expressed comparable levels of native, peptide-receptive cell surface Kb molecules, but generated cell surface Kb-OVA257-264 complexes at least 20-fold less efficiently from ER-targeted peptides. The inefficient assembly of Kb-OVA257-264 complexes in the ER of insect cells cannot be attributed solely to a requirement for human tapasin, since first, human cells lacking tapasin expressed endogenously synthesized Kb- OVA257-264 complexes at levels comparable to tapasin-expressing cells, and second, vaccinia virus-mediated expression of human tapasin in insect cells did not detectably enhance the expression of Kb-OVA257-264 complexes. The assembly of Kb-OVA257-264 complexes could be greatly enhanced in insect but not human cells by a nonproteasomal protease **inhibitor**. These findings indicate that insect cells lack one or more factors required for the efficient assembly of class I-peptide complexes in vertebrate cells and are consistent with the idea that the missing component acts to protect antigenic peptides or their immediate precursors from degradation.

L38 ANSWER 2 OF 3 EMBASE COPYRIGHT 2002 ELSEVIER SCI. B.V.

95253444 EMBASE Document No.: 1995253444. Calnexin influences folding of human class I histocompatibility proteins but not their assembly with . **beta.2-microglobulin**. Tector M.; Salter R.D.. Dept. of Pathology, 733 Scaife Hall, Pittsburgh Univ. School of Medicine,Pittsburgh, PA 15261, United States. Journal of Biological Chemistry 270/33 (19638-19642) 1995. ISSN: 0021-9258. CODEN: JBCHA3. Pub. Country: United States. Language: English. Summary Language: English.

AB Class I major histocompatibility complex heavy chains bind to calnexin before associating with **.beta.2-microglobulin** (.beta.2m) and peptides. Calnexin has been shown to retain in the endoplasmic reticulum those class I heavy chains which have not assembled properly and, thus, to serve as a quality control mechanism. In addition, calnexin may direct the folding of class I subunits or their subsequent assembly. We asked whether calnexin plays a role in the initial folding of HLA-B*0702 heavy chains by assessing disulfide bond formation in vivo. Our results show that class I heavy chains form intrachain disulfide bonds very soon after translation, and that calnexin is bound to both reduced and oxidized forms during this process. When a cell-permeable reducing agent, dithiothreitol, was added to cells, disulfide bond formation in newly synthesized heavy chains was substantially blocked, as was their association with calnexin. The reducing agent appeared to affect calnexin directly, since binding was similarly abolished to a subset of proteins which do not contain internal disulfide bonds. Addition of the glucosidase **inhibitor** castanospermine to cells, shown previously to disrupt calnexin binding to ligands, slowed formation of disulfide bonds but did not decrease the amount of assembled heavy chain-.beta.2m complexes that formed. Our data suggest that calnexin can promote disulfide bond formation in class I heavy chains but does not directly facilitate subsequent binding to .beta.2m.

L38 ANSWER 3 OF 3 EMBASE COPYRIGHT 2002 ELSEVIER SCI. B.V.

94329777 EMBASE Document No.: 1994329777. Phosphatase **inhibitors** block in vivo binding of peptides to class I major histocompatibility complex molecules. Tector M.; Zhang Q.; Salter R.D.. Dept. of Pathology, 733 Scaife Hall, Pittsburgh Univ. School of Medicine,Pittsburgh, PA 15261, United States. Journal of Biological Chemistry 269/41 (25816-25822) 1994.
ISSN: 0021-9258. CODEN: JBCHA3. Pub. Country: United States. Language: English. Summary Language: English.

AB Class I major histocompatibility complex (MHC) molecules are heterotrimers of heavy chains, **.beta.2-microglobulin**, and 8-10 amino acid-long peptides. Assembly of class I MHC molecules into complexes which are stable and can be transported to the cell surface occurs soon after insertion of individual subunits into the endoplasmic reticulum (ER). To identify subcellular compartments required for class I MHC assembly, we studied class I biosynthesis in human cell lines treated with several **inhibitors** of intracellular transport. We found that HLA-B701 molecules do not assemble in CIR transfectants in which a block in protein transport from the ER is established by treatment with phosphatase **inhibitors**. In contrast, stable HLA-B701 complexes form in cells in which the ER becomes mixed with the Golgi after treatment with brefeldin A. Neither treatment impaired binding of HLA- B701 to the ER-resident protein calnexin, and unassembled heavy chains in phosphatase-inhibited cells showed prolonged association with calnexin. In addition, the mouse class I molecule H-2Db, which binds **.beta.2-microglobulin** in human T2 cells in the absence of transporter of antigenic peptides, formed complexes in CIR cell transfectants treated with phosphatase **inhibitors**. Taken together, these data demonstrate that phosphatase **inhibitors** do not prevent assembly of class I heavy chain-**.beta.2-microglobulin** dimers, but instead interfere with peptide loading. These results are consistent with the possibility that class I MHC molecules are transported from their initial site of insertion into the rough ER before binding peptides, or alternatively that peptide loading mediated by transporter of antigenic peptides is blocked by phosphatase **inhibitors**.

```
=> s (stevens f?/au or argon y?/au or davis d?/au)
L39      13130 (STEVENS F?/AU OR ARGON Y?/AU OR DAVIS D?/AU)

=> s l39 and protein aggregation
L40         16 L39 AND PROTEIN AGGREGATION

=> dup remove l40
PROCESSING COMPLETED FOR L40
L41          10 DUP REMOVE L40 (6 DUPLICATES REMOVED)

=> d l41 1-10 cbib abs
```

L41 ANSWER 1 OF 10 EMBASE COPYRIGHT 2002 ELSEVIER SCI. B.V.DUPLICATE 1
2001412377 EMBASE Both the environment and somatic mutations govern the aggregation pathway of pathogenic immunoglobulin light chain. **Davis D.P.**; Gallo G.; Vogen S.M.; Dul J.L.; Sciarretta K.L.; Kumar A.; Raffen R.; **Stevens F.J.**; **Argon Y.**. Y. Argon, Department of Pathology, University of Chicago, Chicago, IL 60637, United States. yargon@midway.uchicago.edu. Journal of Molecular Biology 313/5 (1021-1034) 9 Nov 2001.
Refs: 52.
ISSN: 0022-2836. CODEN: JMOBAK. Pub. Country: United Kingdom. Language: English. Summary Language: English.

AB Deposition of monoclonal immunoglobulin light chain (LC) aggregates in tissues is the hallmark of a class of fatal diseases with no effective treatment. In the most prevalent diseases two different types of LC

aggregates are observed: Fibrillar deposits in LC amyloidosis (AL) and granular aggregates in LC deposition disease (LCDD). The mechanisms by which a given LC forms either type of aggregate are not understood. Although some LCs are more aggregation-prone than others, this does not appear to be due to specific sequence determinants, but more likely from global properties that can be introduced by multiple somatic mutations. Moreover, a single LC isotype can sometimes form both fibrillar and granular aggregates within the same patient. To better understand how the different aggregation pathways arise, we developed a series of in vitro assays to analyze the formation of distinct aggregate types. The recombinant .kappa.IV LC (SMA) assembles into fibrils when agitated. We now show that SMA can also form granular aggregates upon exposure to copper, and that this aggregation can occur not only in vitro, but also in cells. A constellation of somatic mutations, consisting of His89/His94/Gln96, is sufficient to confer sensitivity to copper on wild-type .kappa.IV proteins. The formation of both types of aggregates is inhibited by synthetic peptides derived from the LC variable domain. However, the peptide that inhibits fibrillar aggregation is different from the peptide that inhibits copper-induced aggregation. Thus, distinct molecular surfaces of the LC underly each type of aggregate. We conclude that both the intrinsic properties of the sequence and extrinsic conditions govern the aggregation pathway of a LC. .COPYRGT. 2001 Academic Press.

L41 ANSWER 2 OF 10 EMBASE COPYRIGHT 2002 ELSEVIER SCI. B.V.

2002128866 EMBASE Hsp70 and antifibrillogenic peptides promote degradation and inhibit intracellular aggregation of amyloidogenic light chains. Dul J.L.; **Davis D.P.**; Williamson E.K.; **Stevens F.J.**; **Argon Y.**. Y. Argon, Department of Pathology, University of Chicago, MC 1089, 5841 South Maryland Ave., Chicago, IL 60637, United States. yargon@midway.uchicago.edu. Journal of Cell Biology 152/4 (705-715) 19 Feb 2001.
Refs: 42.
ISSN: 0021-9525. CODEN: JCLBA3. Pub. Country: United States. Language: English. Summary Language: English.

AB In light chain (LC) amyloidosis an immunoglobulin LC assembles into fibrils that are deposited in various tissues. Little is known about how these fibrils form in vivo. We previously showed that a known amyloidogenic LC, SMA, can give rise to amyloid fibrils in vitro when a segment of one of its .beta. sheets undergoes a conformational change, exposing an Hsp70 binding site. To examine SMA aggregation in vivo, we expressed it and its wild-type counterpart, LEN, in COS cells. While LEN is rapidly oxidized and subsequently secreted, newly synthesized SMA remains in the reduced state. Most SMA molecules are dislocated out of the ER into the cytosol, where they are ubiquitinylated and degraded by proteasomes. A parallel pathway for molecules that are not degraded is condensation into perinuclear aggresomes that are surrounded by vimentin-containing intermediate filaments and are dependent upon intact microtubules. Inhibition of proteasome activity shifts the balance toward aggresome formation. Intracellular aggregation is decreased and targeting to proteasomes improved by overexpression of the cytosolic chaperone Hsp70. Importantly, transduction into the cell of an Hsp70 target peptide, derived from the LC sequence, also reduces aggresome formation and increases SMA degradation. These results demonstrate that an amyloidogenic LC can aggregate intracellularly despite the common presentation of extracellular aggregates, and that a similar molecular surface mediates both in vitro fibril formation and in vivo aggregation. Furthermore, rationally designed peptides can be used to suppress this aggregation and may provide a feasible therapeutic approach.

L41 ANSWER 3 OF 10 CAPLUS COPYRIGHT 2002 ACS

2000:323243 Document No. 132:331667 Method using pretreatment and size-exclusion chromatography for detecting and diagnosing disease caused

by pathological **protein aggregation**. **Stevens, Fred J.**; Myatt, Elizabeth A.; Solomon, Alan (United States Dept. of Energy, USA; University of Tennessee). U.S. US 6063636 A 20000516, 10 pp., Division of U.S. Ser. No. 282,473, abandoned. (English). CODEN: USXXAM. APPLICATION: US 1996-605294 19960119. PRIORITY: US 1994-282473 19940729.

AB A method is provided for detecting pathol. macromols. in a patient, comprising obtaining body fluid from the patient, pretreating the body fluid, subjecting the pretreated body fluid to size-exclusion chromatog. to create an excluded fluid, and analyzing the excluded fluid to detect macromols. having a predetd. mol. wt. The method also allows for comparing elution spectra with ref. spectra of suspect pathol. proteins. The aggregation propensity of more than 50 different Bence-Jones proteins were tested under conditions of pH, salt, and urea concn. that would mimic those found within the nephron. HPLC was performed using Superose-12.

L41 ANSWER 4 OF 10 BIOSIS COPYRIGHT 2002 BIOLOGICAL ABSTRACTS INC.

2000:345601 Document No.: PREV200000345601. Small zone, high-speed gel filtration chromatography to detect **protein aggregation** associated with light chain pathologies. Raffen, Rosemarie (1); **Stevens, Fred J.**. (1) Biosciences Division, Argonne National Laboratory, Argonne, IL, 60439 USA. Wetzel, Ronald. Methods in Enzymology, (1999) Vol. 309, pp. 318-332. Methods in Enzymology; Amyloid, prions, and other protein aggregates. print. Publisher: Academic Press Inc. 525 B Street, Suite 1900, San Diego, CA, 92101-4495, USA. ISSN: 0076-6879. ISBN: 0-12-182210-9 (cloth). Language: English. Summary Language: English.

L41 ANSWER 5 OF 10 BIOSIS COPYRIGHT 2002 BIOLOGICAL ABSTRACTS INC.DUPLICATE 2

1999:194337 Document No.: PREV199900194337. Physicochemical consequences of amino acid variations that contribute to fibril formation by immunoglobulin light chains. Raffen, Rosemarie; Dieckman, Lynda J.; Szpunar, Meredith; Wunschl, Christine; Pokkuluri, Phani R.; Dave, Poras; Stevens, Priscilla Wilkins; Cai, Xiaoyin; Schiffer, Marianne; **Stevens, Fred J. (1)**. (1) Argonne National Laboratory, 9700 S. Cass Ave., Building 202, A-141, Argonne, IL, 60439 USA. Protein Science, (March, 1999) Vol. 8, No. 3, pp. 509-517. ISSN: 0961-8368. Language: English.

AB The most common form of systemic amyloidosis originates from antibody light chains. The large number of amino acid variations that distinguish amyloidogenic from nonamyloidogenic light chain proteins has impeded our understanding of the structural basis of light-chain fibril formation. Moreover, even among the subset of human light chains that are amyloidogenic, many primary structure differences are found. We compared the thermodynamic stabilities of two recombinant kappa4 light-chain variable domains (VLs) derived from amyloidogenic light chains with a VL from a benign light chain. The amyloidogenic VLs were significantly less stable than the benign VL. Furthermore, only the amyloidogenic VLs formed fibrils under native conditions in an in vitro fibril formation assay. We used site-directed mutagenesis to examine the consequences of individual amino acid substitutions found in the amyloidogenic VLs on stability and fibril formation capability. Both stabilizing and destabilizing mutations were found; however, only destabilizing mutations induced fibril formation in vitro. We found that fibril formation by the benign VL could be induced by low concentrations of a denaturant. This indicates that there are no structural or sequence-specific features of the benign VL that are incompatible with fibril formation, other than its greater stability. These studies demonstrate that the VL beta-domain structure is vulnerable to destabilizing mutations at a number of sites, including complementarity determining regions (CDRs), and that loss of variable domain stability is a major driving force in fibril formation.

L41 ANSWER 6 OF 10 EMBASE COPYRIGHT 2002 ELSEVIER SCI. B.V.

1999353412 EMBASE Pathogenic light chains and the B-cell repertoire.
Stevens F.J.; Argon Y.. Y. Argon, Dept. of Pathology, Committee on Immunology, University of Chicago, Chicago, IL 60637, United States. yargon@midway.uchicago.edu. Immunology Today 20/10 (451-457) 1999.
Refs: 46.
ISSN: 0167-5699. CODEN: IMTOD8.
Publisher Ident.: S 0167-5699(99)01502-9. Pub. Country: United Kingdom.
Language: English.

L41 ANSWER 7 OF 10 MEDLINE DUPLICATE 3
1999436921 Document Number: 99436921. PubMed ID: 10507033. Small zone, high-speed gel filtration chromatography to detect **protein aggregation** associated with light chain pathologies. Raffen R; **Stevens F J**. (Biosciences Division, Argonne National Laboratory, Illinois 60439, USA.) METHODS IN ENZYMOLOGY, (1999) 309 318-32. Journal code: 0212271. ISSN: 0076-6879. Pub. country: United States. Language: English.
AB Small zone gel filtration chromatography can be used for qualitative and quantitative analysis of protein interactions and aggregation phenomena. The technique is fast, accessible to most laboratories, and can be combined with computer simulation to extract quantitative information from experimental data. The programs KRUNCH and SCIMZ will be furnished on written request to the authors.

L41 ANSWER 8 OF 10 EMBASE COPYRIGHT 2002 ELSEVIER SCI. B.V.
97341164 EMBASE Document No.: 1997341164. Interaction between glycosaminoglycans and immunoglobulin light chains. Jiang X.; Myatt E.; Lykos P.; **Stevens F.J.**. F.J. Stevens, CMBB, Argonne National Laboratory, Argonne, IL 60439, United States. Biochemistry 36/43 (13187-13194) 1997.
Refs: 23.
ISSN: 0006-2960. CODEN: BICHAW. Pub. Country: United States. Language: English. Summary Language: English.
AB Amyloidosis is a pathological process in which normally soluble proteins polymerize to form insoluble fibrils (amyloid). Amyloid formation is found in a number of diseases, including Alzheimer's disease, adult-onset diabetes, and light-chain-associated amyloidosis. No pharmaceutical methods currently exist to prevent this process or to remove the fibrils from tissue. The search for treatment and prevention methods is hampered by a limited understanding of the biophysical basis of amyloid formation. Glycosaminoglycans (GAGs) are long, unbranched heteropolysaccharides composed of repeating disaccharide subunits and are known to associate with amyloid fibrils. The interaction of amyloid-associated free light chains with GAGs was tested by both size-exclusion high-performance liquid chromatography and sodium dodecyl sulfate-polyacrylamide gel electrophoresis experiments. The results indicated that heparin 16 000 and chondroitin sulfate B and C precipitated both human intact light chains and recombinant light chain variable domains. Although all light chains interacted with heparin, the strongest interactions were obtained with proteins that had formed amyloid. Molecular modeling indicated the possibility of interaction between heparin and the conserved saddlelike surface of the light chain dimer opposite the complementarity-determining segments that form part of the antigen-binding site of a functional antibody. This suggestion might offer a new path to block the aggregation of amyloid-associated light chain proteins, by design of antagonists based on properties of GAG binding. A hexasaccharide was modeled as the basis for a possible antagonist.

L41 ANSWER 9 OF 10 CAPLUS COPYRIGHT 2002 ACS
1995:706839 Document No. 123:137872 A fluorescence study of Nile Red bound to human serum albumin in buffer, denaturant, and reverse micelles. **Davis, Daniel M.**; Birch, David J. S.; Gellert, Paul R.; Kittlety,

Rodney S.; Swart, Ronald M. (Department Physics and Applied Physics, University Strathclyde, Glasgow, G4 ONG, UK). Proc. SPIE-Int. Soc. Opt. Eng., 2388(Advances in Fluorescence Sensing Technology II), 302-13 (English) 1995. CODEN: PSISDG. ISSN: 0277-786X.

AB Nile red non-covalently binds to Human Serum Albumin (HSA) in at least two binding sites with distinctly different accessibilities for acrylamide quenching. Here, the authors report on the fluorescence characteristics of the probe-protein complex in various environments using both steady state and time-resolved single photon counting techniques. In particular, fluorescence depolarization measurements demonstrate that the unfolding of a protein by heat is fundamentally different from that using denaturant, regarding the changes in diffusional rotation of the probe at intermediate stages. The authors also exploit the fluorescence of the probe-protein complex in AOT reverse micelles, to increase the understanding of the nature of compartmentalized biol. mols. The large Stokes shift of Nile red allows the changes in the environment of the probe-protein complex in reverse micelles of varying waterpool size, to be obsd. Moreover, comparison of acrylamide quenching of the tryptophanyl residue and bound Nile red in reverse micelles of varying compn., show that there is an induced stability in the nanosecond motions of the protein in reverse micelles of waterpool diam. 80.ANG.. Both far and near UV CD show that at this waterpool size, the protein structure is nearest it's native state. This waterpool size is about the same size as the central cavity in the mol. chaperone GroEL, which suggests that compartmentalization of proteins "in vivo" aids the protein folding process by inducing stability in the appropriate conformation as well as preventing **protein aggregation**.

L41 ANSWER 10 OF 10 BIOSIS COPYRIGHT 2002 BIOLOGICAL ABSTRACTS INC.
1991:199246 Document No.: BR40:96526. INDUCTION OF **PROTEIN AGGREGATION** IN NEPHROMIMETIC SOLUTIONS. MYATT E A; **STEVENS F J**. ARGONNE NATL. LAB., ARGONNE, ILL. 60439-4833, USA.. THIRTY-FIFTH ANNUAL MEETING OF THE BIOPHYSICAL SOCIETY, SAN FRANCISCO, CALIFORNIA, USA, FEBRUARY 24-28, 1991. BIOPHYS J. (1991) 59 (2 PART 2), 119A. CODEN: BIOJAU. ISSN: 0006-3495. Language: English.

=>

=>
Executing the logoff script...

=> LOG H

COST IN U.S. DOLLARS	SINCE FILE ENTRY	TOTAL SESSION
FULL ESTIMATED COST	203.18	203.39

DISCOUNT AMOUNTS (FOR QUALIFYING ACCOUNTS)	SINCE FILE ENTRY	TOTAL SESSION
CA SUBSCRIBER PRICE	-2.48	-2.48

SESSION WILL BE HELD FOR 60 MINUTES
STN INTERNATIONAL SESSION SUSPENDED AT 16:53:22 ON 18 JUL 2002